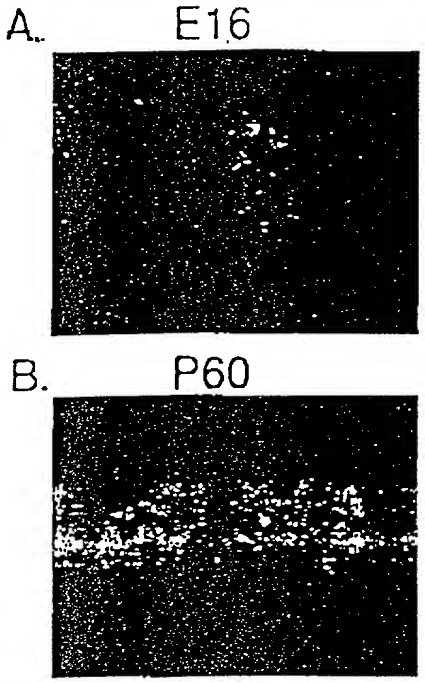
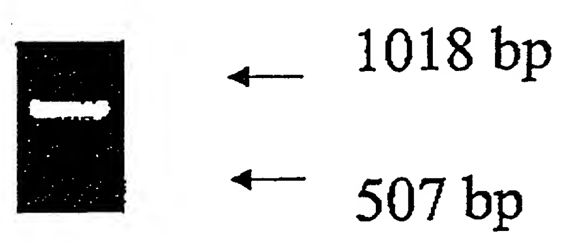


Fig. 1

Figure 2

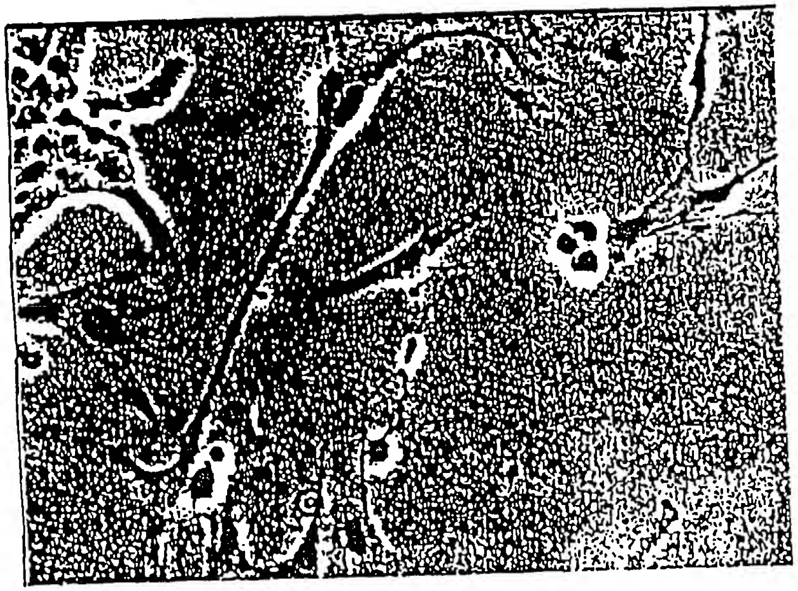
Nestin RT-PCR of 50 rat islets



Amplification of a single band of the correct size of 834 bp. In between the forward [GCGGGGCGGTGCGTGACTAC] and reverse primer [GGGTGGTGAGGGTTGAGGTTTGTG] are 3 introns located.

Figure 3

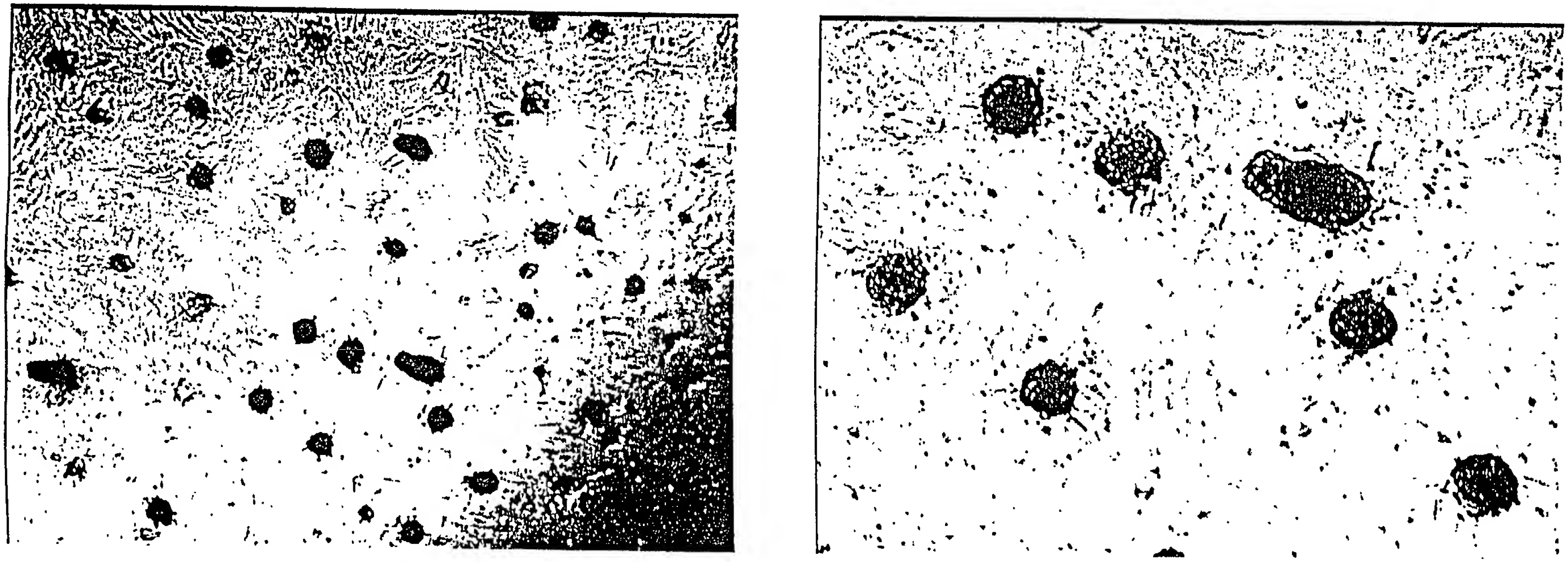
Nestin positive cells proliferate around islets in vitro



Phase contrast image of cells
surrounding cultured islets (200x)

Figure 4

Development of islet-like structures in vitro



100x

200x

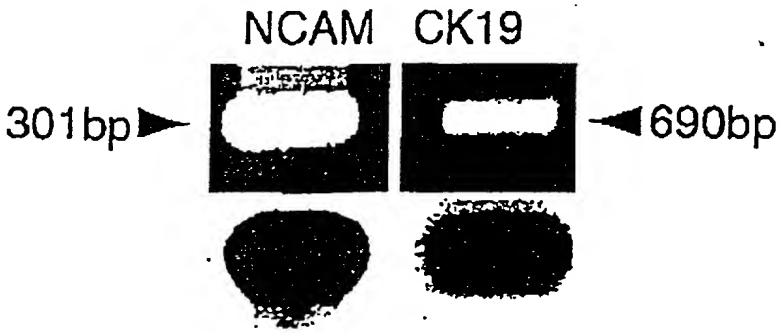
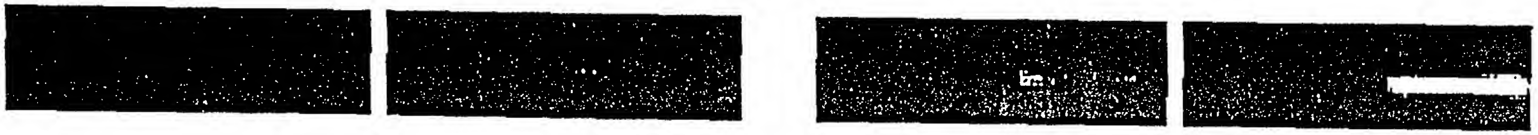


Fig. 5

Figure : 6

Induction of nestin mRNA expression by high glucose in pancreatic islets

RT-PCR of 50 rat islets incubated for 4 days at 5.6 mM or 16.7 mM glucos



	APRT				Nestin			
	low glucose		high glucose		low glucose		high glucose	
cycle	26	27	26	27	32	34	32	34
ratio high/low glucose			1.74	1.69			3.18	2.91

Figure 7 (i

Nestin Amino Acid Sequence:
"MEGCMGEESFQMWELNRRLEAYLGRVKALEEQNELLSAGLGGLR
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ARERTTEEVARNRRAVEAEKCARAWLSSQGAELERELEALRVAHEEERVGLNAQAAC
APRLPAPPRPPAPAPEVEELARRLGEAWRGAVRGYQERVAHMETSLDQTRERLARAVQ
GAR
EVRLELQQLQAERGGLLERRAALEQRLEGRWQERLRATEKFQLAVEALEQEKQGLQSQ
IAQVLEGRQQLAHLKMSLSLEVATYRTLLEAENSRLQTPGGGSKTSLSFQDPKLELQF
PRTPEGRRLGSLLPVLSPTSLPSPLPATLETPVPAFLKNQEFLQARTPTLASTPIPPT
PQAPSPAVDAEIRAQDAPLSLLQTQGGRKQAPEPLRAEARVAIPASVLPGPEEPGGQR
QEASTGQSPEDHASLAPPLSPDHSSLEAKDGESGGSRVFSICRGEGEGQIWGLVEKET
AIEGKVVSSLQQEIWEEEDLNRKEIQDSQVPLEKETLKSLGEEIQESLKTLENQSHET
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SGGNESSRKGNSRTTGVCGSEPRDIQTPGRGESGIIEISGSMEPGEFEISRGVDKESQ
RNLEEEENLGKGEYQESLRSLEEEGQELPQSADVQRWEDTVEKDQELAQESPPGMAGV
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LGSEEAEEDVPPVLVSPSPTYTPILEDAPGLQPQAEGSQEASWGVQGRAEAGKVESEQ
EELGSGEIPEGLQEEGEESREESEEDELGETLPDSTPLGFYLRSPTSPRWTPLESRGH
PLKETGKEGWDPAVLASEGLEEPSEKEEGEEGEEECGRDSDLSEEFEDLGTEAPFLPG
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GSSVGSLQALSSSQRGEFLESDSVSVSVPWDDSLRGAVAGAPKTALETESQDSAEPSG
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GLEQSEESGARNALVSEGDRGSPFQEEEGSALKRSSAGAPVHLGQGQFLKFTQREGDR
ESWSSGED"

Nestin Nucleotide Sequence:
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gcctacctgg gccgggtcaa ggcgctggag gagcagaatg agctgctcag cgccggactc 121
gggggctcc ggcgacaatc cgcggacacc tcctggcggg cgcatgccga cgacgagctg 181
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cgcgacaacc tggctgaaga gctggagggc gtggcaggcc gatgcgagca gctgcggctg 301
gcccgggagc ggacgacgga ggaggtagcc cgcaaccggc gcgccgtcga ggcagagaaa
361 tgcgcccggg cctggctgag tagccagggg gcagagctgg agcgcgagct agaggctcta
421 cgcgtggcgc acgaggagga gcgcgtcggt ctgaacgcgc aggctgcctg tgcccccgc

Figure 7 (continued) (2)

```
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601 acgtcgctgg accagacccg cgagcgcctg gcccgggcgg tgcagggtgc ccgcgaggtc
661 cgcctggagc tgcagcagct ccaggctgag cgcggaggcc tcctggagcg cagggcagcg
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841 ctggaaggtc ggcagcagct ggcgcacctc aagatgtccc tcagcctgga ggtggccacg
901 tacaggaccc tcctggaggc tgagaactcc cggctgcaaa cacctggcgg tggctccaag
961 acttccctca gctttcagga ccccaagctg gagctgcaat tccctaggac cccagagggc
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1141 acccctacct tggccagcac ccccatcccc cccacacctc aggcaccctc tcctgctgta
1201 gatgcagaga tcagagccca ggatgctcct ctctctctgc tccagacaca gggtgggagg
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Figure 7 (Continued) (3)

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ggtggggacc ccaggcctcc aggctcccca ggggctgcca 3181 gaggcgatag agcccctggt
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4441 gccctggaaa cggagtccca ggacagtgct gagccttctg gctcagagga agagtctgac
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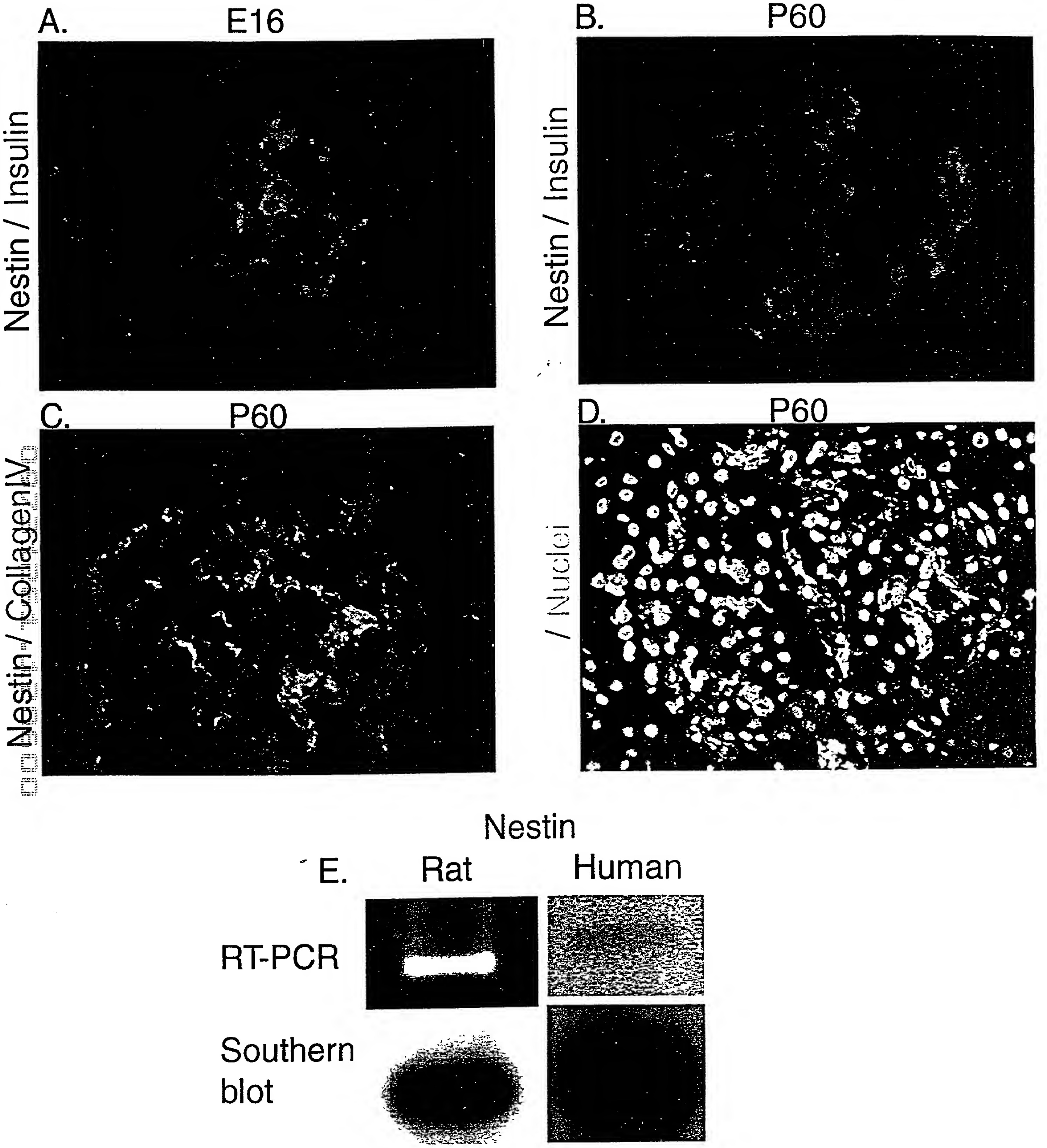

A.
E16
Nestin / Insulin
B.
P60
Nestin / Insulin
C.
P60
Nestin / CollagenIV
D.
P60
/ Nuclei
Nestin
E.
Rat
Human
RT-PCR
Southern blot

Fig 8

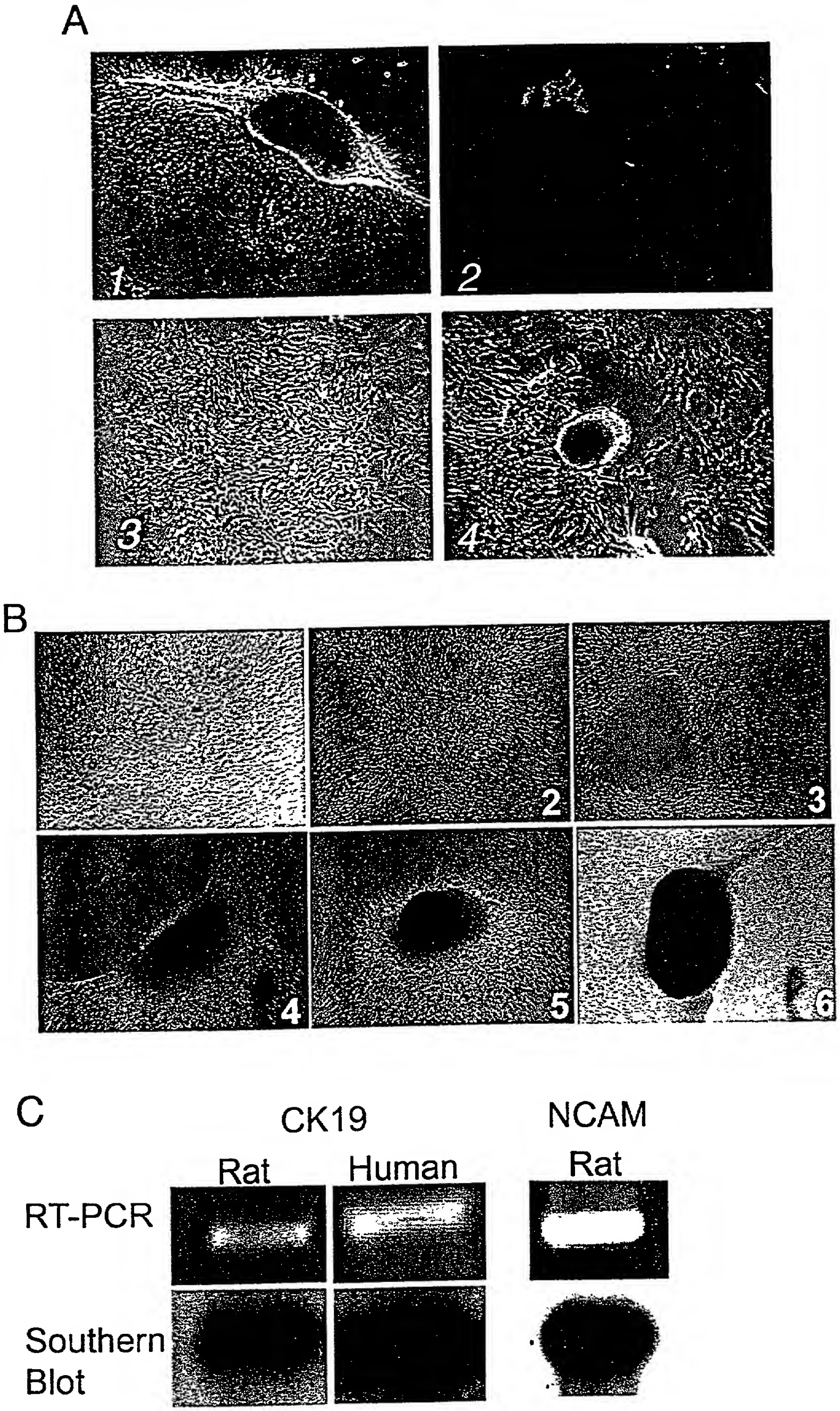


FIG 9

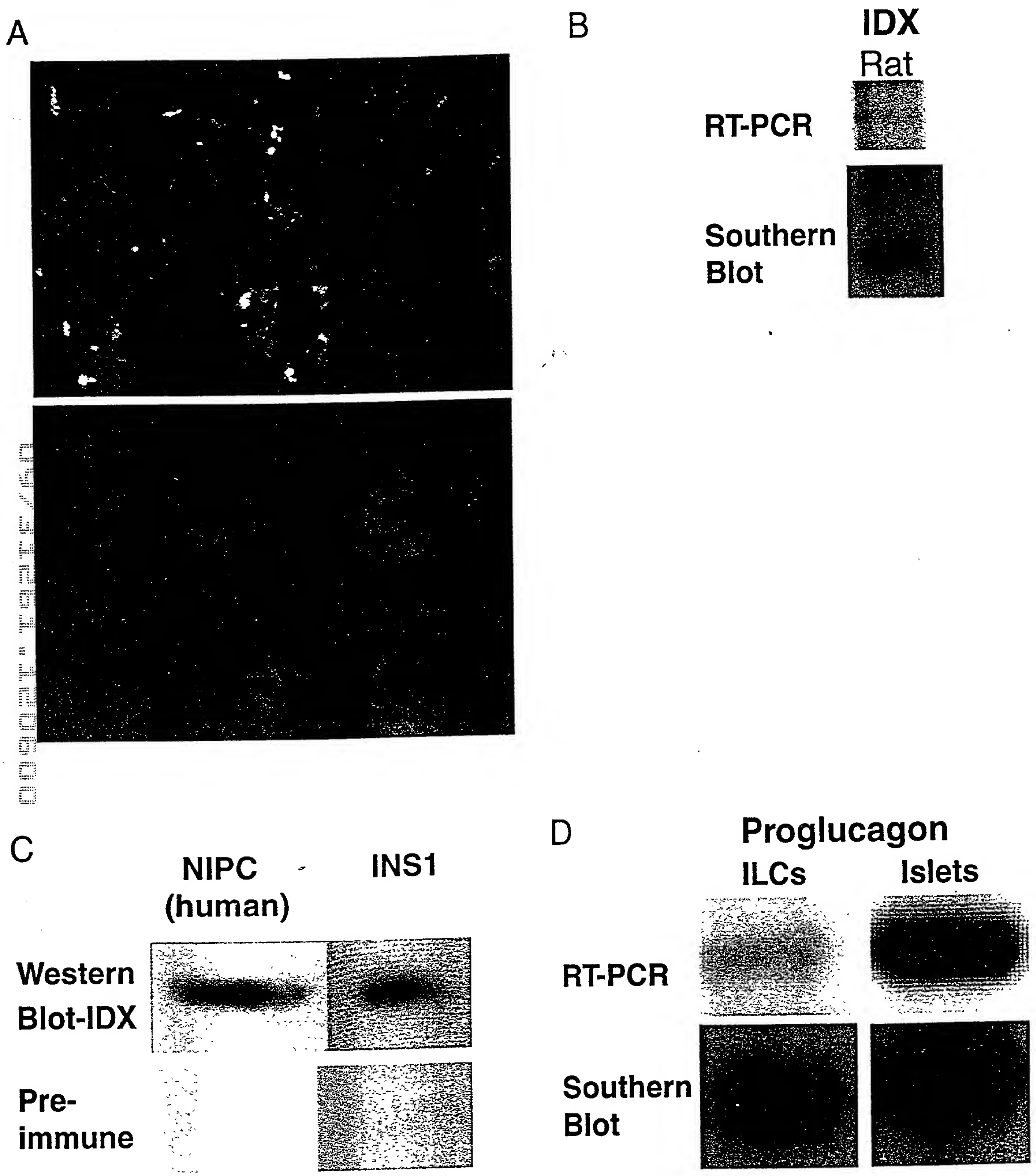

A
B
IDX
Rat
RT-PCR
Southern Blot
C
NIPC (human)
INS1
Western Blot-IDX
Pre-immune
D
Proglucagon
ILCs
Islets
RT-PCR
Southern Blot

FIG 10

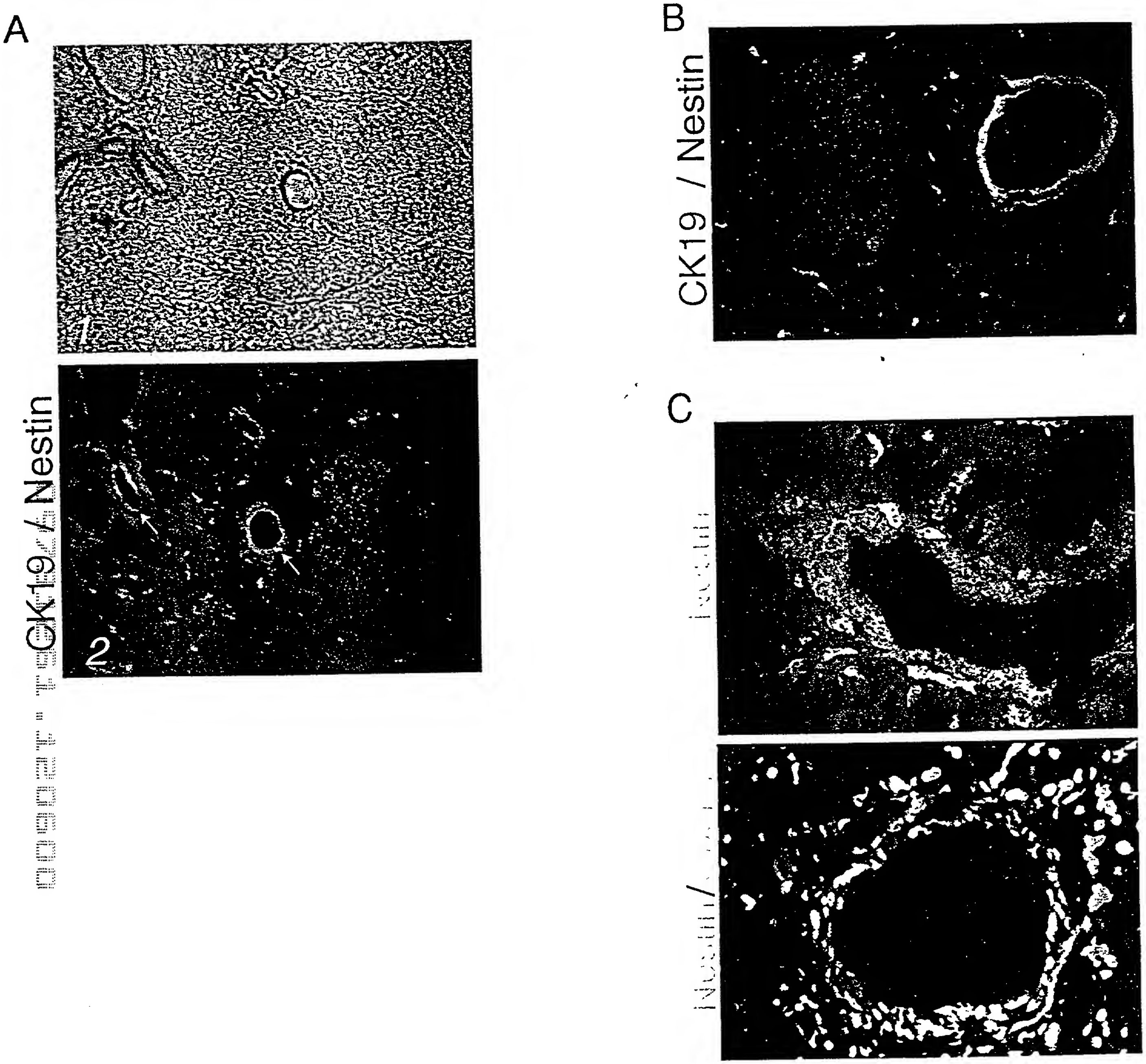
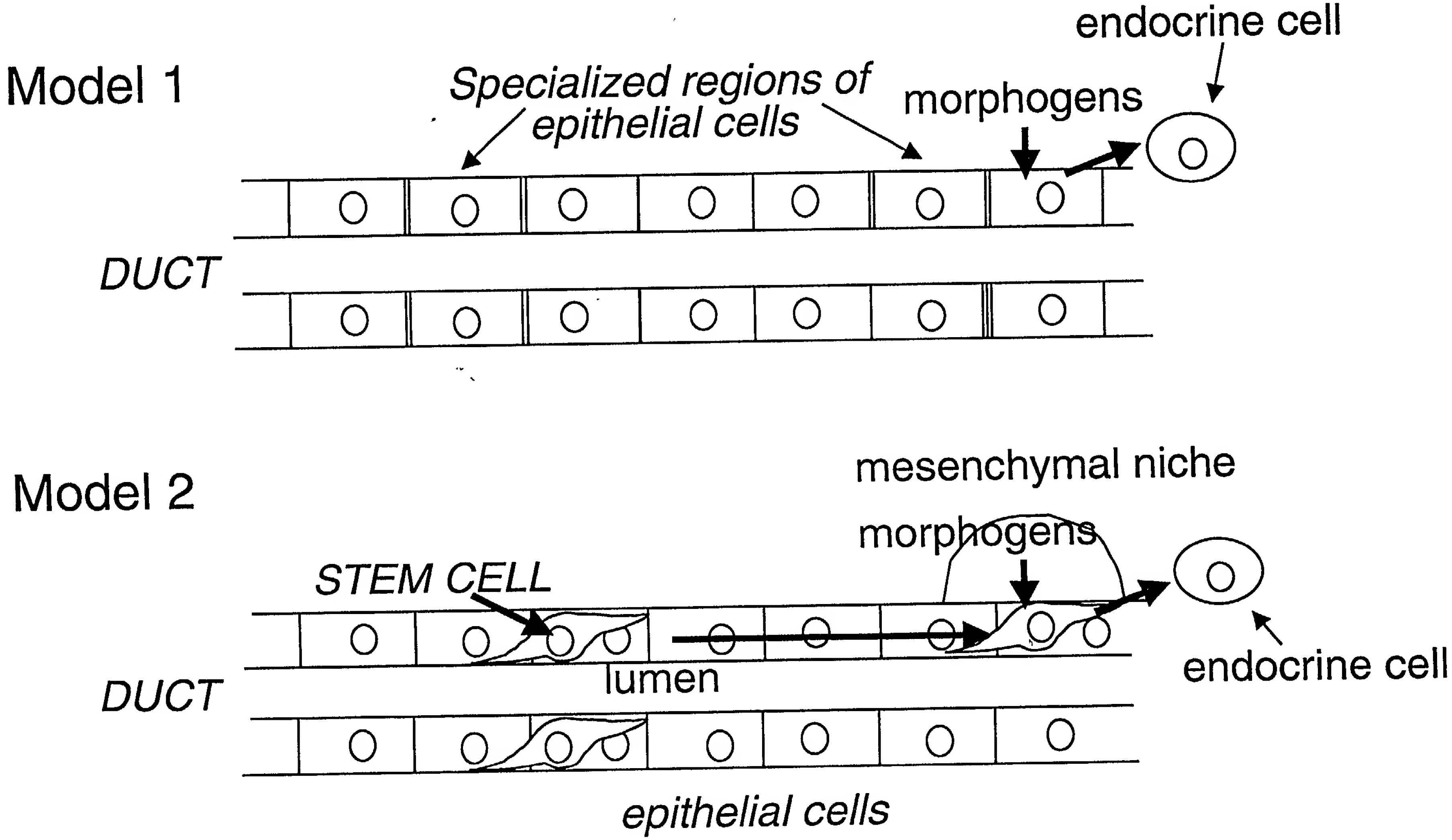


FIG 11

FIG 12
FIG 12



Zulewski et al., Fig.

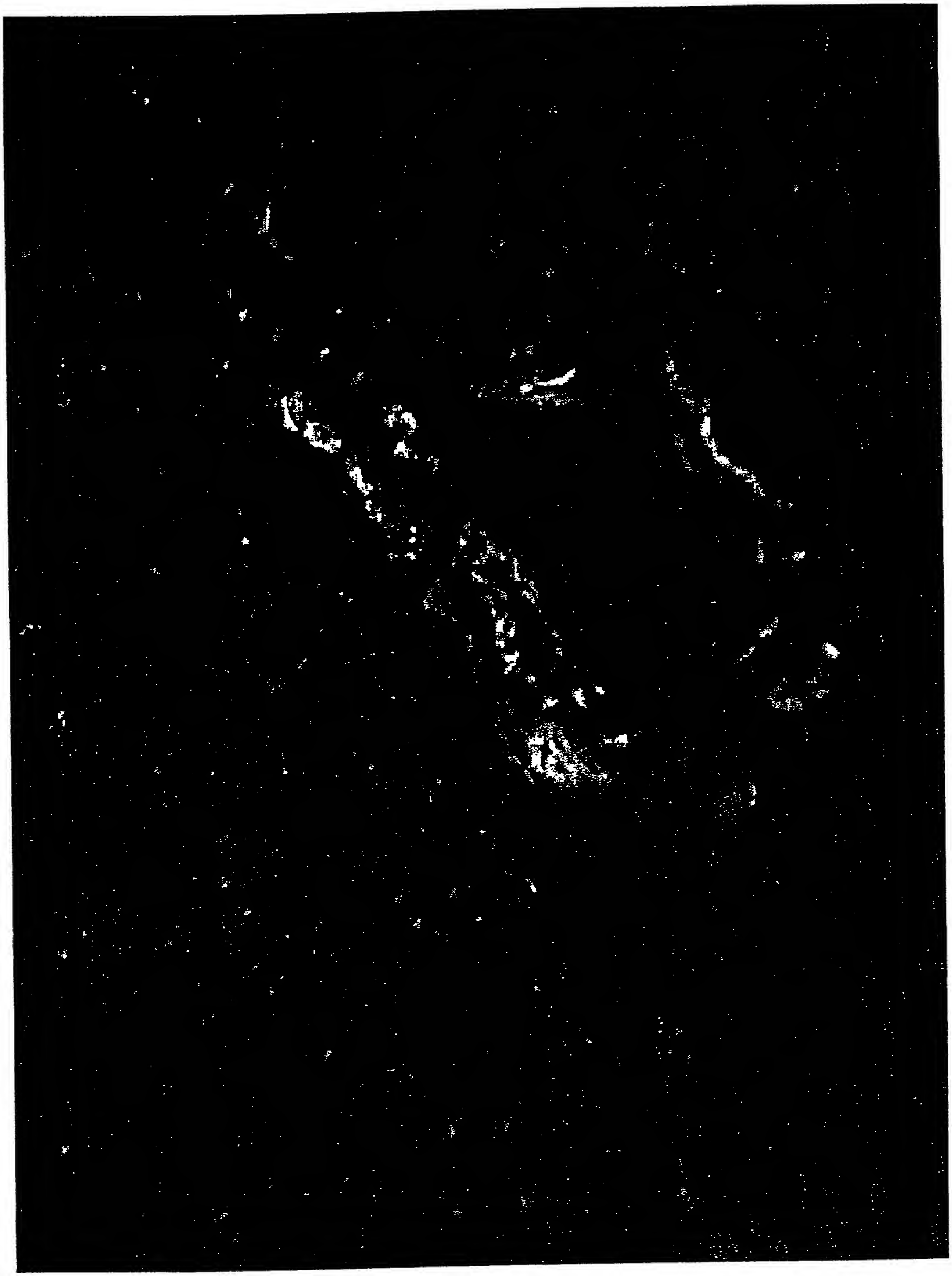


FIG 13A

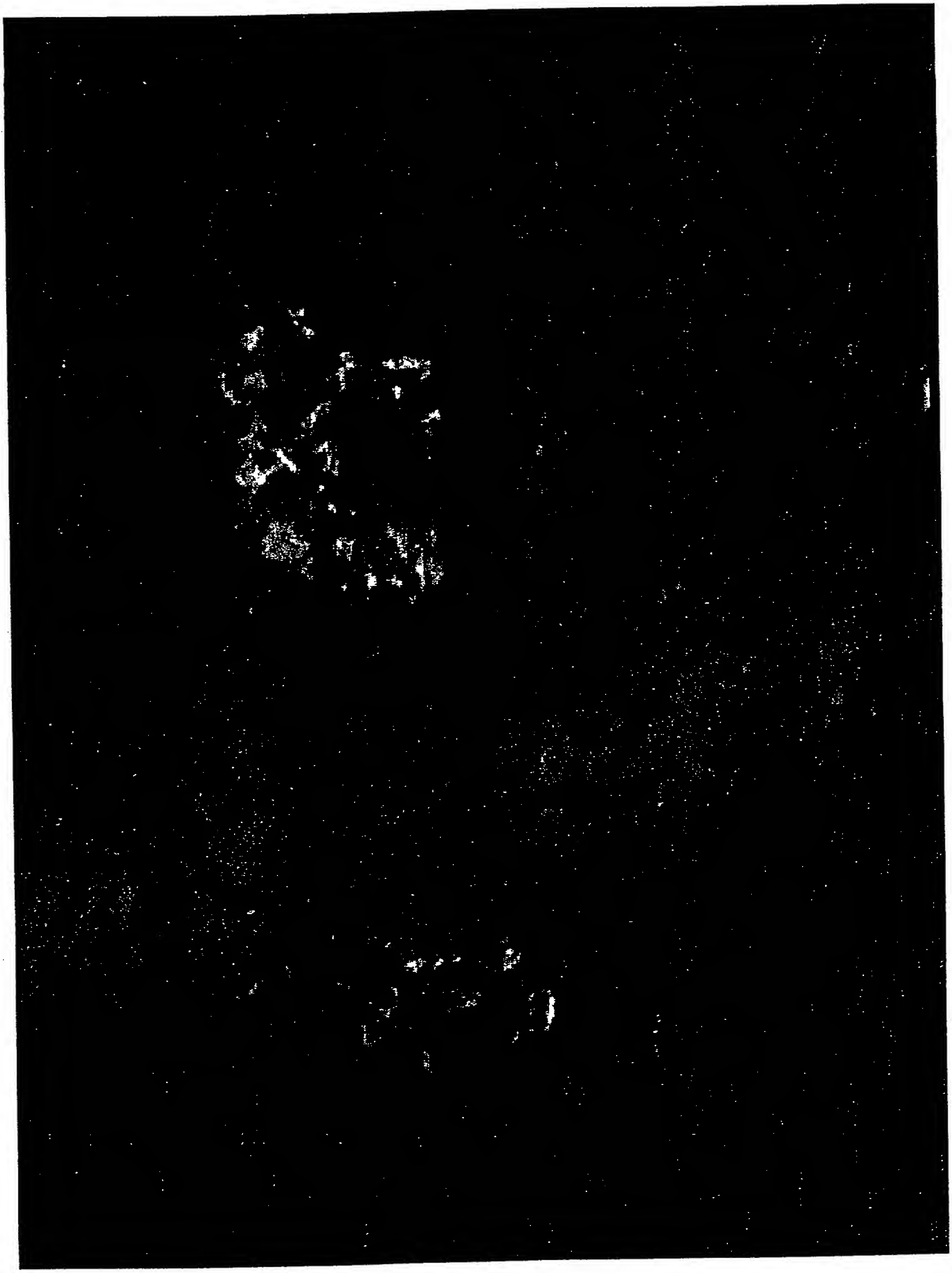
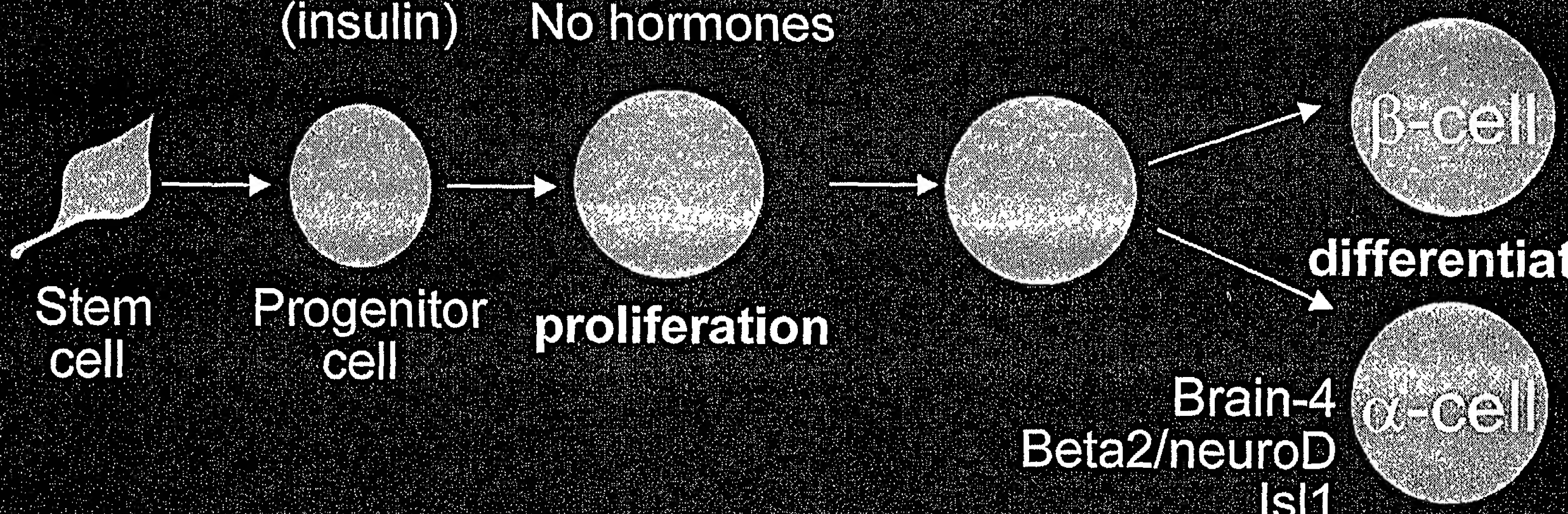


FIG 13 B

Sequential appearance of transcription factors during development of the endocrine pancreas (mouse)



Day: E8.5 E9.5 E13 E14 E15

1st transition

2nd transition

PDX-1
glucagon
(insulin)

PDX-1
Ngn-3
No hormones

PDX-1
Beta2/neuroD
Isl1
Pax6
Nkx6.1

Stem cell

Progenitor cell

proliferation

differentiation

Brain-4
Beta2/neuroD
Isl1
Pax6

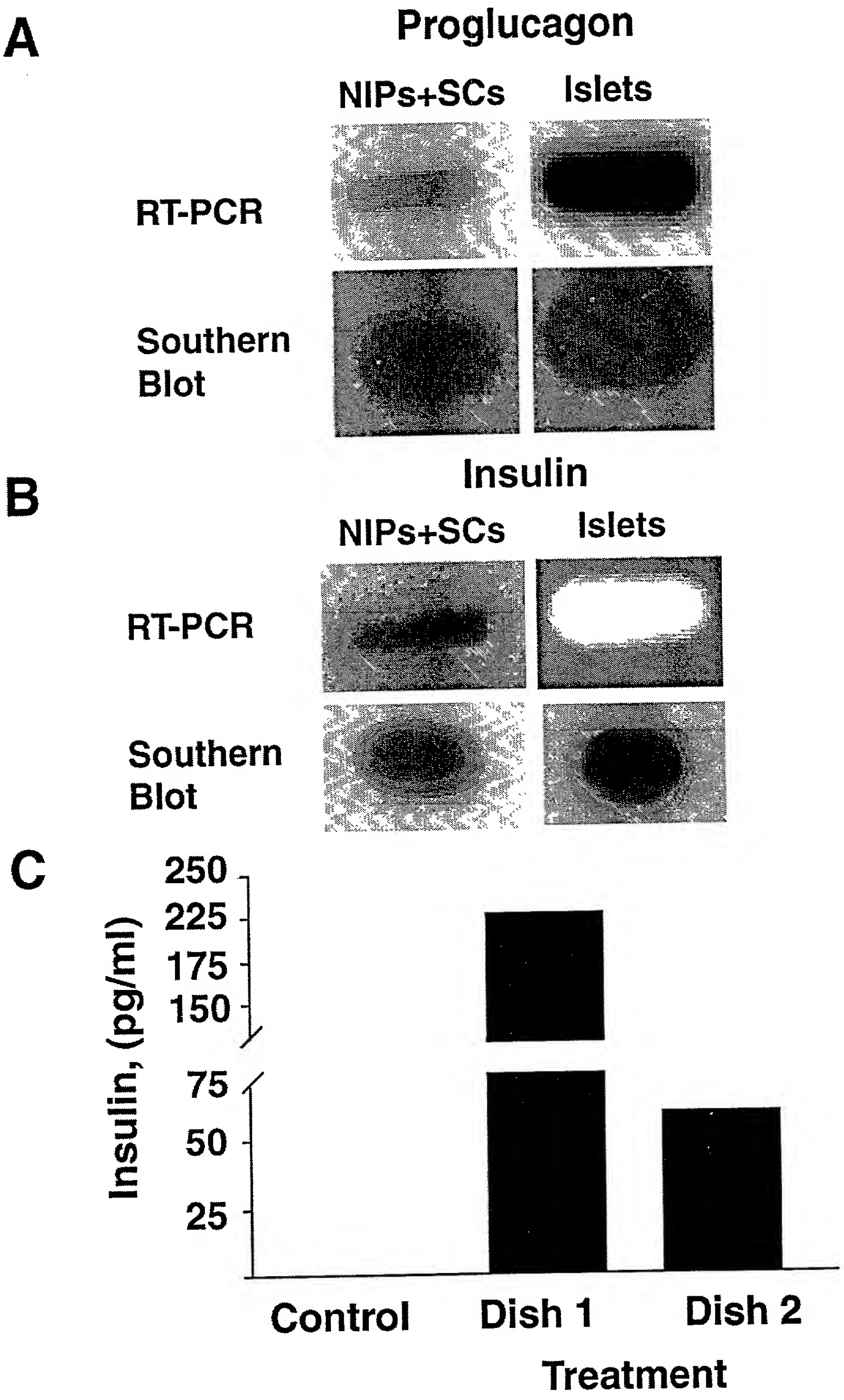

A
Proglucagon
NIPs+SCs
Islets
RT-PCR
Southern Blot
B
Insulin
NIPs+SCs
Islets
RT-PCR
Southern Blot
C
250
225
175
150
75
50
25
Insulin, (pg/ml)
Control
Dish 1
Dish 2
Treatment

FIG 15

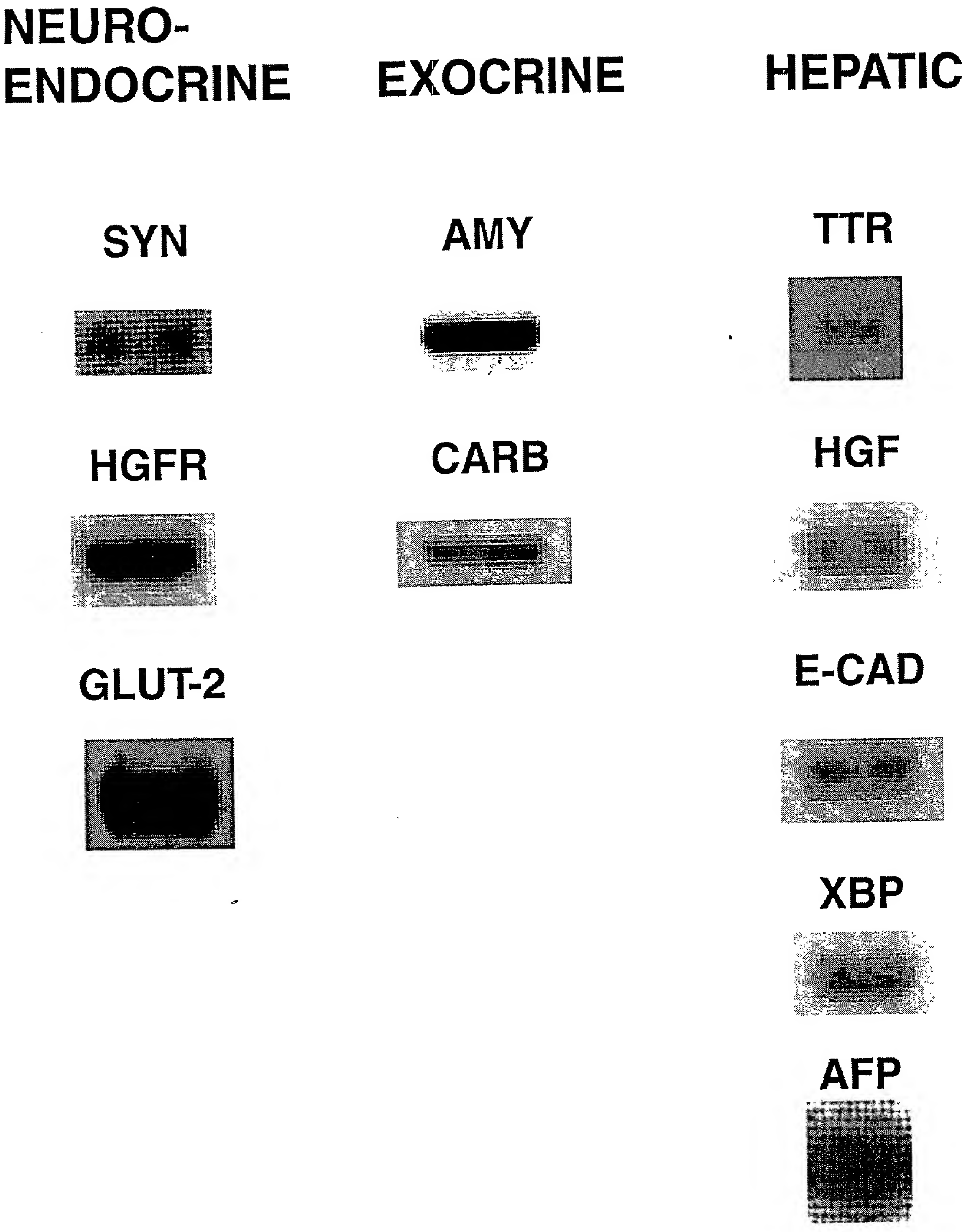

NEURO-
ENDOCRINE
EXOCRINE
HEPATIC
SYN
AMY
TTR
HGFR
CARB
HGF
GLUT-2
E-CAD
XBP
AFP

Fig 16